Amendments to the Claims

Complete Listing of the Claims

1. (Currently amended) A method of characterizing single circulating epithelial cancer cells obtained from about 5 mL to 75 mL of blood comprising:

concurrently measuring multiple cellular markers using fluorescent probes, wherein said probes emit different wavelengths of light to distinguish multiple cellular markers expressed in said cells using fluorescence microscopy, wherein said fluorescent probes comprise five fluorescent probes.

2. (Currently amended) The method of claim 1, wherein said cells are isolated by density gradient centrifugation from a sample containing cells, said isolated cells are adhered onto a surface and fixed with a fixative solution, and said surface containing said cells for characterization is incubated with said probes, wherein each probe reacts with a marker of ~~the single~~ said cells, and any probe binding with a marker is examined by a microscope equipped with an optical filter set for identification of each specific marker.

Claims 3-8 (Cancelled)

9. (Previously presented) The method of claim 2, wherein the surface for cell adherence is a microscope slide.

10. (Previously presented) The method of claim 2, wherein the fixative is selected from a group consisting of paraformaldehyde, formaldehyde, alcohol, or acetone.

11. (Currently amended) The method of claim 1, wherein one or more of said probes is covalently linked to a fluorescent compound that emits a wavelength of light to create a fluorescent probe that binds to a cellular marker.

12. (Currently amended) The method of claim 11, wherein one or more of said fluorescent probes is selected from other probes with minimal overlapping emission spectra for concurrent use in characterizing said cells.

13. (Original) The method of claim 12, wherein said fluorescent probes are selected from a group consisting of a mixture of fluorescent probes that emit light of wavelengths between 400 nanometers and 850 nanometers, wherein said emission spectra can be distinguished from each other with the use of a microscope equipped with spectral filters that allow for elimination of most overlapping wavelengths of fluorescent light being emitted by each selected probe.

14. (Currently amended) The method of claim 1, wherein one or more of said fluorescent probes emits light with wavelengths between 430 nanometers to 510 nanometers.

15. (Currently amended) The method of claim 14, wherein one or more of said fluorescent probes emits light with a peak wavelength of about 470 nanometers.

16. (Currently amended) The method of claim 1, wherein one or more of said fluorescent probes emits light with wavelengths between 482 nanometers to 562 nanometers.

17. (Currently amended) The method of claim 16, wherein one or more of said fluorescent probes emits light with a peak wavelength of about 522 nanometers.

18. (Currently amended) The method of claim 1, wherein one or more of said fluorescent probes emits light with wavelengths between 552 nanometers to 582 nanometers.

19. (Currently amended) The method of claim 18, wherein one or more of said fluorescent probes emits light with a peak wavelength of about 567 nanometers.

20. (Currently amended) The method of claim 1, wherein one or more of said fluorescent probes emits light with wavelengths between 577 nanometers to 657 nanometers.

21. (Currently amended) The method of claim 20, wherein one or more of said fluorescent probes emits light with a peak wavelength of about 617 nanometers.

22. (Currently amended) The method of claim 1, wherein one or more of said fluorescent probes emits light with wavelengths between 637 nanometers to 697 nanometers.

23. (Currently amended) The method of claim 22, wherein one or more of said fluorescent probes emits light with a peak wavelength of about 667 nanometers.

24. (Currently amended) The method of claim 1, wherein one or more of said fluorescent probes emits light with wavelengths between 730 nanometers to 814 nanometers.

25. (Currently amended) The method of claim 24, wherein one or more of said fluorescent probes emits light with a peak wavelength of about 772 nanometers.

26. (Cancelled)

27. (Cancelled)

28. (Currently amended) The method of claim 13, wherein one or more of said fluorescent ~~compounds~~ probes are selected from a group consisting of fluorescein isothiocyanate; CY3; CY3.5; CY5; CY5.5; AMCA; Tetramethylrhodamine Isothiocyanate; TEXAS RED™; R-Phycoerythrin; and Spectral Red.

29. (Cancelled)

30. (Cancelled)

31. (Original) The method of claim 1, wherein the probes comprise 6 fluorescent probes.

32. (Original) The method of claim 1, wherein the probes comprise 7 fluorescent probes.

33. (Original) The method of claim 1, wherein the probes comprise multiple fluorescent probes that emit light of different wavelengths with minimal interference between the wavelengths of emitted light when using appropriate filter set combinations that allow one marker to be distinguished from another when tested concurrently.

34. (Currently amended) The method of claim 1, wherein one or more of said probes is directed to a cellular target and is not a nucleic acid.

35. (Currently amended) The method of claim 34, wherein one or more of said probes comprises a protein or peptide.

36. (Currently amended) The method of claim 35, wherein one or more of said probes is an antibody.

37. (Currently amended) The method of claim 1, wherein one or more of said probes is a nucleic acid directed to a cellular target.

38. (Currently amended) The method of claim 37, wherein one or more of said probes comprises DNA.

39. (Currently amended) The method of claim 37, wherein one or more of said probes comprises RNA.

40. (Previously presented) The method of claim 1, wherein said probes comprise

(i) probes which are directed to a cellular target and are not a nucleic acid,
(ii) probes which are a nucleic acid directed to a cellular target, or
(iii) a combination of (i) and (ii).

41. (Previously presented) The method of claim 40, wherein said probes are selected from the group consisting of identification probes, proliferation probes, cell cycle arrest probes, oncogenes, and hormonal probes.

42. (Cancelled)

43. (Currently amended) The method of claim 40, wherein said probes ~~comprises~~ comprise an epithelial cell-specific probe.

44. (Previously presented) The method of claim 40, wherein the probes comprise a tissue-specific probe.

45. (Currently amended) The method of claim 1, wherein said cells ~~is~~ are obtained from a mammal.

46. (Original) The method of claim 45, wherein said mammal is a human.

47. (Previously presented) The method of claim 40, wherein said probes are used to detect a hormone receptor or a hormone receptor gene for the enumeration of copy number.

48. (Currently amended) The method of claim 47, wherein said hormone receptor or hormone receptor gene is an androgen receptor or androgen receptor gene.

49. (Currently amended) The method of claim 47, wherein said hormone receptor or hormone receptor gene is an estrogen receptor or estrogen receptor gene.

50. (Currently amended) The method of claim 47, wherein said hormone receptor or hormone receptor gene is a progesterone receptor or hormone receptor gene.

51. (Currently amended) The method of claim 1, wherein one or more of said cellular markers is an antigen.

52. (Currently amended) The method of claim 51, wherein one or more of said cellular markers is a receptor.

53. (Currently amended) A method of characterizing ~~a~~ single circulating epithelial cancer cells ~~preparation~~ obtained from about 5 to 75 ml of blood, said method comprising adhering ~~a~~ circulating epithelial cancer cells ~~preparation~~ to be characterized onto a surface, fixing said cell preparation with a fixative solution, incubating said cell surface containing fixed cells with multiple probes directed to desired cellular markers, wherein said multiple probes have the ability to fluoresce when excited at different wavelengths, and examining the cells by fluorescence microscopy for identification of positive cells for each selected cellular marker by concurrent measurement of multiple cellular markers, wherein said cancer cells ~~preparation is~~ are isolated from a body fluid using a negative selection process, wherein said circulating epithelial cancer cells ~~is~~ are obtained, wherein said fluorescent probes comprise five fluorescent probes.

54. (Currently amended) A method of establishing a characterization profile of a circulating epithelial cancer cell obtained from about 5 to 75 ml of blood comprising characterizing a single cell environment by concurrent measurement of multiple cellular markers using fluorescent probes, wherein said probes emit different wavelengths of

light to distinguish multiple cellular markers expressed in the single cell using fluorescence microscopy, wherein said fluorescent probes comprise five fluorescent probes.

55. (Currently amended) The method of claim 53, wherein said ~~single cell is~~ epithelial cancer cells are isolated by density gradient centrifugation from a sample containing cells, said isolated cells are adhered onto a surface and fixed with a fixative solution, and said surface containing cells for characterization is incubated with said probes, wherein each probe reacts with a marker of the single cell, and any probe binding with a marker is examined by a microscope equipped with an optical filter set for identification of each individual marker.

56. (Currently amended) The method of claim 2, wherein said isolated cells are further isolated by a negative selection process.

57. (Cancelled)

58. (Previously presented) The method of claim 2, wherein cells are further isolated by a positive selection process, wherein a specific cell type is selected from a heterogeneous mixture of cells by an antibody that selectively binds to the specific cell type.

59. (Currently amended) The method of any one of claims 1, 53 and 54, wherein one or more of said circulating epithelial cancer cells is a prostatic cancer cell.

60. (Currently amended) The method of any one of claims 1, 53 and 54, wherein one or more of said circulating epithelial cancer cells is a breast cancer cell.

61. (Currently amended) The method of any one of claims 1, 53 and 54, wherein one or more of said circulating epithelial cancer cells is selected from the group

consisting of liver, kidney, colon, rectum, gastric, esophageal, bladder, brain, ovary, pancreas and lung cancer cells.

62. (Cancelled)

63. (Cancelled)

64. (Currently amended) The method of any one of claims 1, 53 and 54, wherein one or more of said circulating epithelial cancer cells is obtained from about 5 to 25 ml of blood.

65. (Currently amended) The method of any one of claims 1, 53 and 54, wherein one or more of said circulating epithelial cancer cells is obtained from about 15 to 25 ml of venous blood.

66. (Currently amended) The method of any one of claims 1, 53 and 54, wherein one or more of said circulating epithelial cancer cells is obtained from about 20 ml of blood.

67. (Cancelled)

68. (Currently amended) The method of claim 64, wherein one or more of said circulating epithelial cancer cells is a prostatic cancer cell.

69. (Currently amended) The method of claim 65, wherein one or more of said circulating epithelial cancer cells is a prostatic cancer cell.

70. (Currently amended) The method of any one of claims 1, 53 and 54, wherein said probes are selected from the group consisting of:

(a) tissue specific probes for determining the cellular origin of the cell;

(b) probes specific ~~cell~~ for tumor cell markers;

(c) probes specific for aneuploidy;

(d) probes specific for cellular markers of proliferation;

(e) probes specific for cellular markers of cell growth inhibition;

(f) probes specific for cell cycle arrest;

(g) probes specific for cellular markers of apoptosis; and

(h) probes specific for hormonal receptors.